EFFECTIVENESS OF COMBINING FLOTATION AND STAINING TECHNIQUES WHEN SORTING BENTHIC INVERTEBRATES[1]

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ABSTRACT: Several methods for quickly and precisely separating benthic organisms from collected substrate have been suggested. We tested the effectiveness of using a flotation technique versus a combination of flotation and staining techniques. The flotation method required less time than combining flotation and staining techniques, but failed to adequately recover annelids. Consequently, when knowledge of the contribution to diversity of annelids or other dense invertebrates is required, use of a combination of flotation and staining techniques is advisable.

An important requirement for studying benthic invertebrates is an accurate and efficient method for sorting organisms. Many techniques have been advanced to decrease sorting time while ensuring the retention of captured taxa. Early methods focused on using saturated sucrose solutions as a flotation medium in which benthic invertebrates were separated from substrate by differences in specific gravity (Anderson 1959, Flannagan 1973, Merickel 1978). Residual sediments were then sorted by hand for invertebrates with high specific gravities. More recent studies have suggested using a combination of techniques, mostly flotation (either sucrose or NaCl) and staining (Thorp and Covich 1991, Wetzel and Likens 1991). In our analysis of the benthic fauna of playa lakes, we found a NaCl solution in conjunction with staining to be a superior technique, especially in the recovery of annelids.

MATERIALS AND METHODS

The benthic fauna of ten playa lakes was surveyed with a 2" ID, single-core sampler. Two hundred fifty substrate samples were taken from each lake and immediately preserved with 10% formalin. In the laboratory, each benthic sample was placed in a wash bucket and immersed in a supersaturated NaCl solution. Floating material was collected with a hand strainer and preserved in 80% ethyl alcohol. The remaining (non-floating) benthic material subsequently was removed from the NaCl solution and transferred from the wash

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bucket into a white enamel sorting pan. Tap water was added to the pan until the sediment was covered by a thin layer of water. Approximately one gm of rose bengal stain was added to the sediment and mixed thoroughly. The mixture was allowed to stand for 30 min then returned to the wash bucket where it was thoroughly rinsed with tap water. The washed sediment was then transferred into a clean white enamel pan for sorting by hand.

RESULTS AND DISCUSSION

Retention and removal of collected organisms using the combination of flotation and staining techniques was superior to flotation alone (Table 1). Using the flotation technique alone, one entire family of annelids (Lumbriculidae) was not detected. Moreover, six times as many leeches (Erpobdellidae) were recovered using the flotation/stain combination technique rather than flotation alone. A G-test of independence (Sokal and Rohlf 1981) revealed significant differences ($P = 0.009$) in families retrieved using the flotation technique versus the flotation/stain technique. However, when the

Table 1. Number of individuals of each family recovered using the flotation method alone versus a combination flotation/staining technique based on 2500 samples divided equally among 10 playas. Alphabetic superscripts denote those families combined for the G-test. Asterisks denote those families whose presence was probably accidental and not used in the G-test.

	Technique	
Benthic Invertebrate Families	**Float alone**	**Float/Stain**
Lumbriculidae[A]	0	5
Erpobdellidae[A]	7	41
Planorbidae[B]	6	22
Carabidae[C]	1	1
Curculionidae[D]	7	7
Scarabidae[C]	3	3
Hydrophilidae[C]	1	2
Chironomidae[E]	8	9
Coenagrionidae[F]	4	4
Aculeata*	1	1
Leptoceridae*	1	1
Caenestheriidae[G]	2	3
Cyprididae[G]	1	1

annelids (Lumbriculidae and Erpobdellidae) were removed from the analysis, no significant differences were found between the two techniques ($P = 0.333$). Therefore, when surveying benthic invertebrates, both flotation and and staining techniques should be used to ensure the detection of all collected organisms. However, if annelids are not a concern, the flotation method is more efficient than the combination technique because substrate staining requires approximately an additional 45 min per sample.

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